

Rearing the Olive Fly Parasitoid *Psytalia concolor*

R. Rodriguez, C. H. Pickett, and M. Robertson¹

Both natural and artificial rearing systems are being developed for the rearing of olive fruit fly (OLFF) parasitoids. Cultures of parasitoids are needed for host testing studies and for release into several sites. We began rearing OLFF in September 2002 using olives collected in the field. The flies reared easily, the only limitation being the seasonal availability of olives. Green, immature fruit are best, but they are not available from April to June. We have begun testing the use of fruit stored in a high nitrogen/low temperature environment with help from Hannah Burrack (Department of Entomology) and Bill Biasi (Department of Pomology) at UC Davis. Olives were field collected from roadside trees in Yolo County and divided into eight two-kilogram lots. The olives were then stored for about two months, from January 30 to April 1. Olives rapidly matured from green-purple to black after removal from storage. Olives proved unacceptable for fly production two weeks after removal from storage due to molding of the fruit. The rapid decay of stored fruit limited the production of *Psytalia concolor* to those OLFF larvae that developed quickly and emerged from the olives. Green olives became available for rearing in mid June. The use of green olives increased parasite production due to an increase in the longevity of the fruit. We began rearing *P. concolor* on infested fruit beginning April 9, 2003. Rearing was conducted at around 22°C in our containment facility in south Sacramento. Olives were exposed to flies in a sleeve cage under artificial lighting (14:10, L:D).

The steps taken for producing parasitoids from these olives were:

1. Expose 80 to 100 green olives to OLFF for two to three days for oviposition.
2. Remove olives from OLFF and place on shelves in 0.5-gallon waxed buckets for three days. Buckets had a false bottom of hardware wire, the bottom covered with vermiculite.
3. Half of the olives from above were exposed to *P. concolor* for three days, the remaining half held for maintenance of the fly culture. Olives were held at room conditions on shelves, or in an environmental chamber set at 14:10, L:D, 25°C, and 75% humidity.
4. Parasitoids were collected as they emerged, approximately 22 days later.

A total of 1,190 parasitoids were successfully reared on olives from April 9 through December 29, 2003 with equal success for samples reared on laboratory shelves and in controlled environment chambers (Table 1). A rearing period of 18 weeks produced 633 parasites, with approximately 35 parasites produced per week (Table 2).

Table 1. Parasite production per cage on laboratory shelves versus controlled environment chambers from June 30 to October 31, 2003.

	No. Pupae Recovered	No. Male Parasites	No. Female Parasites	Total Parasites	Pupae/ Olive	Parasites/ Olive	% Parasitation
Shelves	23.27	3.52	3.20	6.729	2.095	0.606	28.91
STDEV	4.8	0.61	0.57	1.1	0.268	0.09	3.67
Chambers	23.04	3.73	3.15	6.88	1.93	0.57	29.89
STDEV	4.64	0.69	0.65	1.15	0.26	0.08	5.43

Table 2. Total parasite production for all cages and conditions June 30 to October 31, 2003.

Olives	Pupae Recovered	No. Adult Parasites	% Parasitism	Pupae/ Olive	Parasites/ Olive
1,068	2,154	633	29	2	0.59

As an alternative to the use of olive, we began rearing *P. concolor* with flies reared on an artificial medium with assistance from Dr. Mark Robertson. A laboratory strain of olive fly from Greece was used, since they rear much better than wild flies on this diet. Dr. Robertson has been sending us fly eggs, which we in turn rear on the artificial medium.

Protocol for rearing OLFF on an artificial diet:

1. Weigh out 250 grams of artificial diet and place in foam tray eight inches by four inches.
2. Add fly eggs to 20 milliliters of 0.2% propionic acid.
3. Sprinkle eggs with a 30-mill dropper over diet tray until all liquid has been dispensed.
4. Allow trays to incubate at 25°C and 50% humidity for 10 to 11 days.
5. Wash diet containing larvae through mesh screen with tap water at room temperature to free larvae from diet.
6. Add fresh diet to stinging dish (eight centimeter by one centimeter circular plastic dish with screened lid), add larvae to dish, add 20 ml of propionic acid, and then cover with screened lid.
7. Expose to *P. concolor* for 16 to 24 hours by placing the plastic dish inside a larger cage with at least 50 adult *P. concolor*; make sure that the diet does not dry out, add another 20 ml propionic acid if diet does dry out. The parasitoid cage is held at laboratory conditions 22°C and a 14:10 light:dark regime.
8. Remove larvae from the plastic dish and place into a foam tray as above, with fresh diet; and add 20 ml of propionic acid.
9. Place the foam tray into a large Tupperware® container that closes tightly, and has been lined with paper towels. Then place the container back into an environmental chamber set at 25°C and 50% relative humidity.
10. Larvae will begin to pupate in the next two to three days and will crawl out of the diet tray and under the paper towels.
11. Collect parasitoids as they emerge 12 to 14 days later.

Three cohorts of OLFF larvae were reared on the artificial diet for 10 to 11 days prior to *P. concolor* exposure (Table 3). A total of 452 adult *P. concolor* were collected from the three cohorts four weeks sooner than parasites produced by larvae reared on olives (Table 4).

Table 3. Summary data for *P. concolor* produced from larvae reared on artificial diet.

Cohort	Date of Exposure	Number of Parasites Collected	Number of Puparia Collected	% Parasitism
1	December 17-23	95	336	28
2	January 21-22	278	704	39.4
3	March 1-2	79	186	42
Total	6 days of exposure	452	1,226	36.8

Table 4. Comparison of *P. concolor* rearing on olives versus artificial diet.

	Parasitoids Produced	No. Puparia Collected	% Parasitism	Weeks Reared	Parasites per Week
Olives	633	2,154	29	18	35
Artificial Diet	452	1,226	36.8	14	32

In summary, both rearing techniques produced about the same number of parasitoids per week. However, the artificial diet has more potential for producing a higher number of parasitoids, and most importantly, it does this without the need for fresh olives. Given more fly eggs, we could produce far more parasitoids. This system requires far less space as well. Furthermore, average parasitism was higher when using hosts reared on the artificial diet.

¹ University of California, Riverside, CA.